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APPLICATION NO.	FILING DATE	FIRST NAMED INVENTOR	ATTORNEY DOCKET NO.
09/341,660	09/15/99	BERRY	3101-18PUS

HM11/0720

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EXAMINER	
FRONDA, C	

ART UNIT	PAPER NUMBER
1652	

DATE MAILED: 07/20/00

Please find below and/or attached an Office communication concerning this application or proceeding.

Commissioner of Patents and Trademarks

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APPLICATION NO.	FILING DATE	FIRST NAMED INVENTOR	ATTORNEY DOCKET NO.
09/341,600	09/15/99	BERRY A	3161-18-PUS

HM12/0526

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EXAMINER	
FRONDA, C	
ART UNIT	**PAPER NUMBER**
1652	7

DATE MAILED: 05/26/00

Please find below and/or attached an Office communication concerning this application or proceeding.

Commissioner of Patents and Trademarks

Office Action Summary

Application No.	Applicant(s)
09/341,600	Berr et al.
Examiner	**Group Art Unit**
Christian L. Fronda	1652

☐ Responsive to communication(s) filed on _____ .

☐ This action is **FINAL.**

☐ Since this application is in condition for allowance except for formal matters, **prosecution as to the merits is closed** in accordance with the practice under *Ex parte Quayle*, 1935 C.D. 11; 453 O.G. 213.

A shortened statutory period for response to this action is set to expire 3 month(s), or thirty days, whichever is longer, from the mailing date of this communication. Failure to respond within the period for response will cause the application to become abandoned. (35 U.S.C. § 133). Extensions of time may be obtained under the provisions of 37 CFR 1.136(a).

Disposition of Claims

☒ Claim(s) *1-39* is/are pending in the application.

Of the above, claim(s) _____ is/are withdrawn from consideration.

☐ Claim(s) _____ is/are allowed.

☒ Claim(s) *1-39* is/are rejected.

☐ Claim(s) _____ is/are objected to.

☐ Claims _____ are subject to restriction or election requirement.

Application Papers

☐ See the attached Notice of Draftsperson's Patent Drawing Review, PTO-948.

☐ The drawing(s) filed on _____ is/are objected to by the Examiner.

☐ The proposed drawing correction, filed on _____ is ☐approved ☐disapproved.

☐ The specification is objected to by the Examiner.

☐ The oath or declaration is objected to by the Examiner.

Priority under 35 U.S.C. § 119

☐ Acknowledgement is made of a claim for foreign priority under 35 U.S.C. § 119(a)-(d).

☐ All ☐ Some* ☐ None of the CERTIFIED copies of the priority documents have been

☐ received.

☐ received in Application No. (Series Code/Serial Number) _____ .

☐ received in this national stage application from the International Bureau (PCT Rule 17.2(a)).

*Certified copies not received: _____ .

☐ Acknowledgement is made of a claim for domestic priority under 35 U.S.C. § 119(e).

Attachment(s)

☒ Notice of References Cited, PTO-892

☒ Information Disclosure Statement(s), PTO-1449, Paper No(s). *5 and 6*

☐ Interview Summary, PTO-413

☐ Notice of Draftsperson's Patent Drawing Review, PTO-948

☐ Notice of Informal Patent Application, PTO-152

--- SEE OFFICE ACTION ON THE FOLLOWING PAGES ---

DETAILED ACTION

Claim Rejections - 35 U.S.C. § 103

1. The following is a quotation of 35 U.S.C. 103(a) which forms the basis for all obviousness rejections set forth in this Office action:

> (a) A patent may not be obtained though the invention is not identically disclosed or described as set forth in section 102 of this title, if the differences between the subject matter sought to be patented and the prior art are such that the subject matter as a whole would have been obvious at the time the invention was made to a person having ordinary skill in the art to which said subject matter pertains. Patentability shall not be negatived by the manner in which the invention was made.

This application currently names joint inventors. In considering patentability of the claims under 35 U.S.C. 103(a), the examiner presumes that the subject matter of the various claims was commonly owned at the time any inventions covered therein were made absent any evidence to the contrary. Applicant is advised of the obligation under 37 CFR 1.56 to point out the inventor and invention dates of each claim that was not commonly owned at the time a later invention was made in order for the examiner to consider the applicability of 35 U.S.C. 103(c) and potential 35 U.S.C. 102(f) or (g) prior art under 35 U.S.C. 103(a).

2. Claims 1-6, 8, 14-16, 18, 21 are rejected under 35 U.S.C. 103(a) as being unpatentable over Dutka-Malen *et al.* in view of O'Shea *et al.*

Dutka-Malen *et al.* teach the *E. coli* glucosamine synthetase gene, a recombinant vector containing transcriptional control sequence and said gene, transformation of *E. coli* host cell with said recombinant vector, culturing of said transformed *E. coli* host cell, and overexpression and purification of the recombinant enzyme (see entire publication). Dutka-Malen *et al.* do not teach a method for producing glucosamine according to claims 1-6, 8, 14-16, 18, 21. O'Shea *et al.* teach a method for isolating glucosamine-6-phosphate and glucosamine using capillary electrophoresis (see entire publication and **Fig. 3**, p. 950).

It would have been obvious to one of ordinary skill in the art at the time the invention was made to produce glucosamine-6-phosphate or glucosamine according to claims 1-6, 8, 14-16, 18, 21 since the rationale, reagents, and process steps are known.

Glucosamine or glucosamine-6-phosphate is expected to be produced by modifying the teachings of Dutka-Malen *et al.* in which the transformed host cell taught by Dutka-Malen *et al.* is contacted with substrates and glucosamine or glucosamine-6-phosphate is isolated from the culture (fermentation) medium or from the microorganism by using the method taught by O'Shea *et al.* Because inherent to the teachings of Dutka-Malen *et al.* is that glucosamine-6-phosphate is

present intracellularly and glucosamine is secreted to the culture medium, recovery of glucosamine is expected to be achieved by using the method taught by O'Shea *et al.* to isolate the products (*cf.* claims 2 and 3). Because methods for lysing *E. coli* cells are well known in the art, intracellular glucosamine-6-phosphate is expected to be obtained by lysing the transformed *E. coli* cells followed by isolation of the product using the method taught by O'Shea *et al.* (*cf.* claim 4). Because phosphatases are commercially available, a further step of dephosphorylating glucosamine-6-phosphate is expected to be achieved by contacting glucosamine-6-phosphate with said phosphatases (*cf.* claim 5). Because Dutka-Malen *et al.* teach by reference to Maniatis *et al.* (see ***Enzyme assay and purification***, p. 288) the culturing of the transformed *E. coli* cells at a temperature of about 28 - 37 ^{0}C with a carbon source from 0.5% - 5% in a flask (fermentor), glucosamine-6-phosphate or glucosamine is expected to be produced in the said culturing conditions and the products recovered by using the method taught by O'Shea *et al.* (*cf.* claim 6). Because methods are well known to increase the cell density of *E. coli* in culture such as culturing in TB (terrific broth), glucosamine-6-phosphate or glucosamine is expected to be produced at a concentration of at least 1 g/L and the products recovered by using the method taught by O'Shea *et al.* (*cf.* claim 9).

One of ordinary skill in the art would be motivated to produce glucosamine-6-phosphate or glucosamine according to claims 1-6, 8, 14-16, 18, 21 because of the need for new cost-effective methods for production of glucosamine which is a well known, important nutritional product. Furthermore, one of ordinary skill in the art would have had a reasonable expectation for success because the reagents and process steps for producing glucosamine-6-phosphate or glucosamine according to claims 1-6, 8, 14-16, 18, 21 are known in the art and because of the success of Dutka-Malen *et al.* in producing a genetically modified microorganism which overexpresses glucosamine-6-phosphate synthase.

3. Claims 7 and 17 are rejected under 35 U.S.C. 103(a) as being unpatentable over Plumbridge in view of Joyce *et al.* and O'Shea *et al.* Plumbridge teach the following *Escherichia coli* genes which encode enzymes involved in the pathways for amino sugar metabolism: *nagA, nagB, nagC, nagD, nagE, and manXYZ* (see p. 2629). Plumbridge do not teach a genetic modification wherein said genetic modification is a mutation in an *Escherichia coli* gene according to claims 7 and 17. Joyce *et al.* teach a method to delete specific genes from the *Escherichia coli* genome and the successful use of this method to delete the *polA* gene from the *Escherichia coli* genome (see entire publication).

It would have been obvious to one of ordinary skill in the art at the time the invention was made to make an *Escherichia coli* cell having a mutation according to claims 7 and 17 for use in the method of claim 1 by deleting any one of the *Escherichia coli* genes taught by

Plumbridge using the method taught by Joyce *et al.* The mutated *Escherichia coli* cell is then used in production of glucosamine or glucosamine-6-phosphate by contacting the said mutated cell with substrates and glucosamine or glucosamine-6-phosphate isolated from the culture (fermentation) medium or from the microorganism by using the method taught by O'Shea *et al.* One of ordinary skill in the art would be motivated to make an *Escherichia coli* having a mutation according to claim 17 because of the need for new cost-effective methods for production of glucosamine which is a well known, important nutritional product. Furthermore, one of ordinary skill in the art would have had a reasonable expectation for success because of the success of Joyce *et al.* in deleting *polA* gene from the *Escherichia coli* genome.

4. Claim 9 is rejected under 35 U.S.C. 103(a) as being unpatentable over Dutka-Malen *et al.* in view of Balbas *et al.* and O'Shea *et al.*

The teachings of Dutka-Malen *et al.* are described above. Dutka-Malen *et al.* do not teach methods for producing glucosamine-6-phosphate or glucosamine according to claim 9 wherein the gene encoding glucosamine synthetase is integrated into the *E. coli* genome. Balbas *et al.* teach a vector for chromosomal integration of cloned DNA into the *E. coli* genome and the successful integrations of the *Vitreoscilla* sp. hemoglobin-encoding gene and the *Photobacterium leiognathi lux* genes into the *E. coli* genome (see entire publication). In addition, Balbas *et al.* teach the advantages of integration of cloned DNA into the genome of a host organism: stability of the cloned gene, and absence of undesirable copy number effects (see fist paragraph of **Introduction**, p. 65). The teachings of O'Shea *et al.* are described above.

It would have been obvious to one of ordinary skill in the art at the time the invention was made to produce glucosamine or glucosamine-6-phosphate according to claim 9 since the rationale, reagents, and process steps are known.

Glucosamine or glucosamine-6-phosphate is expected to be produced according to claim 9 in which the gene encoding *E. coli* glucosamine synthetase taught by Dutka-Malen *et al.* is cloned into the integrative vector taught by Balbas *et al.* and the recombinant, integrative vector is used to transform an *E. coli* host cell. The transformed *E. coli* host cell is contacted with substrates and glucosamine or glucosamine-6-phosphate is isolated from the culture (fermentation) medium or from the microorganism by using the method taught by O'Shea *et al.* One of ordinary skill in the art would be motivated to clone the gene encoding *E. coli* glucosamine synthetase into the integrative vector and transform an *E. coli* host cell with the recombinant, integrative vector because of the advantages taught by Balbas *et al.* One of ordinary skill in the art would have had a reasonable expectation for success in integrating the gene encoding *E. coli* glucosamine synthetase into the *E. coli* genome because of the success of Balbas *et al.* in successfully integrating the *Vitreoscilla* sp. gene encoding hemoglobin and the

Photobacterium leiognathi lux genes into the *E. coli* genome.

5. Claims 10 and 19 are rejected under 35 U.S.C. 103(a) as being unpatentable over Dutka-Malen *et al.* in view of O'Shea *et al.*

The teachings of Dutka-Malen *et al.* are described above. Dutka-Malen *et al* do not teach a method for producing glucosamine or glucosamine-6-phosphate according to claims 10 and 19. The teachings of O'Shea *et al* are described above. DNA sequences encoding glucosamine-6-phosphate synthase are well known in the art as described above. Methods for mutating DNA such as site-directed mutagenesis, UV irradiation of DNA, and treatment with a mutagenic agent are well known in the art.

It would have been obvious to one of ordinary skill in the art at the time the invention was made to make a genetically modified microorganism according to claims 10 and 19 for use in the method of claim 1 since the rationale, reagents, and process steps for making the microorganism according to claim 10 are known.

Glucosamine or glucosamine-6-phosphate is expected to be produced according to claims 10 and 19 by modifying the teachings of Dutka-Malen *et al.* in which the DNA encoding glucosamine synthetase is mutated by site-directed mutagenesis, UV irradiation, or treatment with mutagenic agent using methods well known in the art; the mutated DNAs are inserted into an expression vector; *E. coli* host cells are transformed by methods well known in the art with said expression vector containing the mutated DNAs; host cells are selected which contain a glucosamine synthetase that has a reduction in product inhibition by assaying for enzyme activity in the presence of product; the selected host cell is used in production of glucosamine or glucosamine-6-phosphate by contacting the selected host cell with substrates and glucosamine or glucosamine-6-phosphate isolated from the culture (fermentation) medium or from the microorganism by using the method taught by O'Shea *et al.*

One of ordinary skill in the art would be motivated to produce glucosamine-6-phosphate or glucosamine according to claims 10 and 19 because of the need for new cost-effective methods for production of glucosamine which is a well known, important nutritional product. Furthermore, one of ordinary skill in the art would have had a reasonable expectation for success because the reagents and process steps for producing glucosamine according to claim 10 is known in the art and because of the success of Dutka-Malen *et al.* in producing a genetically modified microorganism which overexpresses glucosamine synthetase.

6. Claims 11-13 and 20 are rejected under 35 U.S.C. 103(a) as being unpatentable over Dutka-Malen *et al.* in view of Plumbridge, Joyce *et al.*, and O'Shea *et al.*

The teachings of Dutka-Malen *et al.* are described above. Dutka-Malen *et al* do not teach a microorganism according to claims 11-13 and 20. The teachings of Plumbridge, Joyce *et al.*, and O'Shea *et al.* are described above.

It would have been obvious to one of ordinary skill in the art at the time the invention was made to make a microorganism according to claims 11-13 and 20 for use in the method of claim 1 or 18 since the rationale, reagents, and process steps for making the microorganism according to claims 11-13 and 20 are known.

A microorganism according to claims 11-13 and 20 is expected to be produced by further modifying the *E. Coli* host cell described above in the 103 rejection of claims 22, 23, 27-29, and 33 in which any gene or genes taught by Plumbridge is deleted in said host cell by using the method taught by Joyce *et al.* Deletion of any one or combination of the genes taught by Plumbridge is expected to result in a decrease or loss of enzyme activity. This host cell containing a deletion of said gene or genes is then used in production of glucosamine or glucosamine-6-phosphate by contacting the selected host cell with substrates and glucosamine or glucosamine-6-phosphate isolated from the culture (fermentation) medium or from the microorganism by using the method taught by O'Shea *et al.* One of ordinary skill in the art would be motivated to produce a microorganism according to claims 11-13 and 20 for use in the method of claim 1 or 18 because of the need for new cost-effective methods for production of glucosamine which is a well known, important nutritional product.

7. Claims 22, 23, 27-29, and 33 are rejected under 35 U.S.C. 103(a) as being unpatentable over Dutka-Malen *et al.* in view of Balbas *et al.* and O'Shea *et al.*

The teachings of Dutka-Malen *et al.* are described above. Dutka-Malen *et al* do not teach a microorganism according to claims 22, 23, 27-29, and 33. The teachings of Balbas *et al.* are described above. DNA sequences encoding glucosamine-6-phosphate synthase are well known in the art as described above. Methods for mutating DNA such as site-directed mutagenesis, UV irradiation of DNA, and treatment with a mutagenic agent are well known in the art.

It would have been obvious to one of ordinary skill in the art at the time the invention was made to make a microorganism according to claims 22, 23, 27-29, and 33 since the rationale, reagents, and process steps for making the microorganism according to claims 22, 23, 27-29, and 33.

A microorganism according to claims 22, 23, 27-29, and 33 is expected to be produced by modifying the teachings of Dutka-Malen *et al.* in which the DNA encoding glucosamine synthetase is mutated by site-directed mutagenesis, UV irradiation, or treatment with mutagenic

agent using methods well known in the art; the mutated DNAs are inserted into the integrative vector taught by Balbas *et al.* and the recombinant, integrative vector is used to transform an *E. coli* host cell.; and host cells are selected which contain a glucosamine synthetase that has a reduction in product inhibition by assaying for enzyme activity in the presence of product. Because methods are well known to increase the cell density of *E. coli* in culture such as culturing in TB (terrific broth), glucosamine is expected to be produced at a concentration of at least 1 g/L when expression of glucosamine-6-phosphated synthase is induced by 1 mM IPTG and the products recovered by using the method taught by O'Shea *et al.* (*cf* claim 33).

One of ordinary skill in the art would be motivated to produce glucosamine according to claims 22, 23, 27-29, and 33 because of the need for new cost-effective methods for production of glucosamine which is a well known, important nutritional product.

8. Claims 24-26, and 30-32 are rejected under 35 U.S.C. 103(a) as being unpatentable over Dutka-Malen *et al.* in view of Balbas *et al.*, Plumbridge , and Joyce *et al.*

The teachings of Dutka-Malen *et al.* are described above. Dutka-Malen *et al* do not teach a microorganism according to claims 24-26, and 30-32. The teachings of Balbas *et al.*, Plumbridge , and Joyce *et al.* are described above.

It would have been obvious to one of ordinary skill in the art at the time the invention was made to make a microorganism according to claims 24-26, and 30-32 since the rationale, reagents, and process steps for making the microorganism according to claims 24-26, and 30-32.

A microorganism according to claims 24-26, and 30-32 is expected to be produced by further modifying the *E. Coli* host cell described above in the 103 rejection of claims 22, 23, 27-29, and 33 in which any gene taught by Plumbridge is deleted in said host cell by using the method taught by Joyce *et al.* Deletion of any one or combination of the genes taught by Plumbridge is expected to result in a decrease or loss of enzyme activity. One of ordinary skill in the art would be motivated to produce a microorganism according to claims 24-26, and 30-32 because of the need for new cost-effective methods for production of glucosamine which is a well known, important nutritional product.

9. Claims 34-39 are rejected under 35 U.S.C. 103(a) as being unpatentable over Dutka-Malen *et al.* in view of Balbas *et al.*, Plumbridge, Joyce *et al.*, and O'Shea *et al.*

The teachings of Dutka-Malen *et al.* are described above. Dutka-Malen *et al.* do not teach a microorganism according to claim 34-39. The teachings of Balbas *et al.*, Plumbridge, Joyce *et al.*, and O'Shea *et al.* are described above.

It would have been obvious to one of ordinary skill in the art at the time the invention

was made to produce a microorganism according to claims 34-39 since the rationale, reagents, and process steps for making the microorganism are known.

A microorganism is expected to be produced according to claims 34-39 in which the *E. coli* glucosamine synthetase gene taught by Dutka-Malen *et al.* is inserted into the integrative vector taught by Balbas *et al.*; the recombinant, integrative vector is used to transform *E. coli* host cells (*cf.* claim 34); host cells are selected which have increased expression of enzyme activity; and any gene taught by Plumbridge is deleted in said host cell having increased expression of enzyme activity by using the method taught by Joyce *et al.* Deletion of any of the genes taught by Plumbridge is expected to result in a decrease or loss of enzyme activity. Because methods are well known to increase the cell density of *E. coli* in culture such as culturing in TB (terrific broth), glucosamine is expected to be produced at a concentration of at least 1 g/L when expression of glucosamine-6-phosphated synthase is induced by 1 mM IPTG and the products recovered by using the method taught by O'Shea *et al.* (*cf* claim 39).

One of ordinary skill in the art would be motivated to produce a microorganism according to claims 34-39 because of the need for new cost-effective methods for production of glucosamine which is a well known, important nutritional product. Furthermore, one of ordinary skill in the art would have had a reasonable expectation for success because Dutka-Malen *et al.* succeeded in producing a genetically modified microorganism which overexpresses glucosamine-6-phosphate synthase and Joyce *et al.* successfully deleted the *polA* gene from the genome of *Escherichia coli*.

Conclusion

10. No claim is allowed.

Any inquiry concerning this communication or earlier communications from the examiner should be directed to Christian L. Fronda whose telephone number is (703)305-1252. If attempts to reach the examiner by telephone are unsuccessful, the examiner's supervisor, Ponnathapura Achutamurthy, can be reached at (703)308-3804. The fax phone number for this Group is (703)308-0294. Any inquiry of a general nature or relating to the status of this application should be directed to the Group 1600 receptionist whose telephone number is (703)308-0196.

CLF
May 10, 2000

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